STRIDE: A **S**tudy of Patients with Lower Extremity Acu**t**e Limb Ischemia to **R**emove Thrombus with the **I**n**d**igo® Aspiration Syst**e**m

Protocol
CLP-15549.D

Date of Protocol
05/20/2021

Device Name
Indigo® Aspiration System

Sponsor
Penumbra, Inc.
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CLP-15549.D Protocol Synopsis	
Study Title:	**STRIDE: A Study of Patients with Lower Extremity Acute Limb Ischemia to Remove Thrombus with the Indigo® Aspiration System**
Study Objective:	The primary objective of this study is to collect safety and performance data on the Indigo Aspiration System in a patient population with lower extremity acute limb ischemia (LE ALI).
Study Design:	Post-market, real world, prospective, single-arm, multi-center study that will enroll up to 130 subjects at up to 25 sites globally
Indication:	Per Instructions For Use (IFU)
Patient Population:	Patients that present with LE ALI who are eligible for mechanical thrombectomy using the Indigo Aspiration System
Study Device:	Indigo Aspiration System
Study Duration:	It is anticipated that enrollment will take approximately 24 months. Subjects will be in the study for approximately 12 months from enrollment to last follow-up.
Follow-up:	Follow-up assessments will occur at discharge and/or 7 days, 30 days, 180 days and 365 days following the procedure.
Inclusion Criteria:	1. Patient age ≥ 18 2. Patient presents with acute (≤14 days) occlusion of lower limb artery(ies) (below inguinal ligament) 3. Patient with a Rutherford Category I, IIa or IIb score 4. Frontline treatment with Indigo Aspiration System 5. Informed consent is obtained from either patient or legally authorized representative (LAR)
Exclusion Criteria:	1. Life expectancy <1 year 2. Target vessel size <2 mm 3. LE ALI secondary to dissections, vasculitis, and/or target vessel trauma 4. Amputation in the ipsilateral limb 5. Pregnancy or positive pregnancy test according to site specific standards of care (only required for women of childbearing potential, serum or urine acceptable) 6. Absolute contraindication to contrast administration 7. Patient is unwilling or unable to comply with protocol follow up schedule and/or based on the Investigator's judgment the patient is not a good study candidate 8. Currently participating in an investigational drug or device clinical trial that may confound the study endpoints. Patients in observational, natural history, and/or epidemiological studies not involving intervention are eligible. 9. Target thrombus in a saphenous vein bypass graft

CLP-15549.D Protocol Synopsis	
Study Title:	**STRIDE: A Study of Patients with Lower Extremity Acute Limb Ischemia to Remove Thrombus with the Indigo® Aspiration System**
Primary Endpoint:	• Target limb salvage rate at 1-month post-procedure
Secondary Endpoints:	• Technical success defined as TIMI 2/3 flow rate immediate post-procedure • Modified SVS runoff score immediate post-procedure as compared to baseline • Improvement of Rutherford classification of one or more as compared to the pre-procedure • Patency at 1 month • Target limb salvage rate at 12 months post-procedure • Rates of device related serious adverse events (SAEs) • Major bleeding peri-procedure • Mortality at 12 months
Primary Statistical Hypothesis:	The null hypothesis is that the difference between the primary endpoint rate and the standard of care at 1-month post-procedure is less than or equal to -10%. The alternative hypothesis is the difference at 1-month post-procedure is greater than -10%.
Primary Statistical Test:	The primary safety analysis will be the difference between the primary endpoint rate in the study and the combined historical control rate of 95.7% observed under current standards of care with a non-inferiority margin of 10%. The primary safety endpoint is met if the lower limit of the 95% confidence interval of the primary safety endpoint rate is greater than 85.7%.
Sample Size Justification	The sample size calculations assume that 94% (108/115) of the study subjects achieve target limb salvage 1-month post-procedure. Based on a binomial analysis with a non-inferiority margin of 10%, a study of 115 enrolled subjects will have 80% power with a one-sided alpha of 0.025. The sample size was adjusted to 130 subjects to account for approximately 10% attrition.

1 Introduction and Rationale

1.1 Disease State and Prevalence

Acute limb ischemia (ALI) is a vascular emergency characterized by a sudden loss of arterial perfusion in the lower limbs.[1-3] The condition affects an estimated 15-26 patients / 100,000 persons / yr in the United States[1], and is associated with extremely high morbidity and mortality rates. In the UK, a study calculated an annual ALI incidence of 10 in 100,000, with 5-year survival rates for avoiding amputation at 36.7% and overall at 55.9%.[4]

Mortality at 1 yr can reach 40%[1, 5] and post-procedural rates of limb loss are reported in between 12%-50% of cases.[5] Optimal clinical outcomes therefore necessitate urgent recognition of the condition and prompt restoration of arterial blood flow, in order to avoid irreversible ischemia and to support limb salvage.[2, 3]

In ALI, the rapid onset of perfusion limitation (<14 days) threatens limb viability due to insufficient time for adequate collateral recruitment.[6] Clinical diagnosis depends on symptom presentation, which includes the '6P's': pain, pallor, pulse deficit, paresthesia, poikilothermia, and paralysis of the affected limb.[6, 7] Nevertheless, uncertainty remains regarding interventional outcomes for ALI patients, and despite recent improvements in therapeutic technique and declines in 1-yr amputation rate, there have been no significant improvements in 1-yr amputation-free survival, which remains unchanged at 52.3%.[1, 5]

1.2 Current Treatments

Clinical practice recommendations for the diagnosis and management of ALI classify patients using the Rutherford classification scale.[2, 3, 8, 9] Patients falling into Rutherford category I present with no continuing ischemic pain and the limb is not immediately threatened. [8, 10] Patients falling into Rutherford category II (a and b) are at risk of limb loss, with prompt treatment and revascularization urgently indicated for limb salvage.

Contemporary revascularization guidelines for ALI include, but are not limited to, catheter-directed thrombolysis (CDT), percutaneous thrombo-aspiration, and surgical thromboembolectomy.[2, 3] Over the past decades, the surgical management of ALI has evolved to include endovascular approaches,[5, 9] especially following the publication of three randomized, multicenter trials which established the efficacy of catheter-directed thrombolysis (CDT) as compared with surgical revascularization.[10-12] CDT,[13] along with newer endovascular approaches including aspiration thrombectomy, have become routine for restoring perfusion in ALI patients in the Rutherford I and II categories.[3, 5, 10] Nevertheless, the cost of ALI-related complications and mortality remains high, leading to strong interest in assessing the efficacy of different revascularization devices and procedures.[14]

The PEARL registry evaluated 283 patients (mean age 65±13years) from 34 institutions across the US which tracked the use of rheolytic pharmacomechanical thrombectomy (PMT) to manage ALI.[9] This method generates low-pressure suction to remove occlusive lesions. At 12-month follow-up, amputation-free survival and freedom from mortality was 81% and 91%, respectively. This study supported the use of PMT, as a first-line

treatment for ALI, with reduced procedure times and an acceptable risk profile compared with CDT.[9]

Although CDT remains an established technique for ALI treatment, it can be associated with multiple procedures and hemorrhagic complications.[15] A small 2018 study comparing percutaneous aspiration thrombectomy (PAT) (n=15) versus conventional CDT (n=27) in treating noniatrogenic acute lower limb ischemia, found that first-line use of PAT for endovascular treatment of ALI could reduce the need for CDT, without significantly increasing costs.[15]

PRISM, a multicenter retrospective case analysis, was established to determine the safety and initial efficacy of another aspiration–based extraction technique (XTRACT), for treating peripheral arterial thromboembolism using the Penumbra/ Indigo System.[16] Seventy-nine patients (median age 69 years [IQR], 60–78 years) with peripheral occlusions were enrolled treated with XTRACT as the initial therapy or secondary therapy. Complete or near-complete revascularization (TIMI grade 2/3) was achieved in 79.5% of patients who received Indigo as a frontline treatment and successful revascularization was achieved in 92.5% of patients who received aspiration-based therapy as salvage or secondary therapy.[16] Complete flow restoration (TIMI grade 3) was successfully achieved in 77.2% of patients, and no patients required surgical revascularization. No device-related adverse events occurred, establishing the safety and efficacy of using the Penumbra/ Indigo system for aspiration-based extraction of acute peripheral arterial occlusions.[16]

The goal of this study protocol is to create a framework for a prospective, multi-center study to obtain additional clinical evidence for the Indigo System as the frontline PAT approach for ALI patients in the Rutherford I, IIa and IIb categories, for whom immediate treatment and revascularization can maintain limb viability and prevent limb loss. Rapid restoration of arterial flow is crucial. The FRIENDS registry (n=200) showed that delays in treatment of limb ischemia is associated with poorer outcomes in ALI patients, including much higher rates of first amputation ($p=0.0002$) and worse amputation-free survival rates ($p<0.05$).[17] Given the high cost burden and poor clinical outcomes associated with ALI, this registry will provide much-needed clinical data on restoring vessel patency and improving patient morbidity and mortality rates.

2 Device Description

2.1 Indigo® Aspiration System

General Description/Overview

The Indigo Aspiration System is comprised of several devices:

- Indigo Aspiration Catheter
- Penumbra Aspiration Pump
- Indigo Pump/Canister Tubing
- Indigo Aspiration Tubing
- Indigo Separator™

The Indigo Aspiration System is designed to remove thrombus from the vasculature using continuous aspiration. The Indigo Aspiration Catheter targets aspiration from the pump directly to the thrombus. The Indigo Separator may be used to clear the lumen of the Indigo Aspiration Catheter should it become blocked with thrombus. The use of the Indigo Separator may not be necessary when using an Indigo Aspiration Catheter with an I.D. of 0.054 in [1.37 mm] or larger. The Indigo Aspiration Catheter is introduced through a guide catheter or introducer sheath and into the peripheral vasculature and guided over a guidewire to the site of the primary occlusion. The Indigo Aspiration Catheter is used with the Penumbra Aspiration Pump to aspirate thrombus from an occluded vessel. As needed, an Indigo Separator may be deployed from the Indigo Aspiration Catheter to assist with thrombus removal. The Indigo Separator is advanced and retracted through the Indigo Aspiration Catheter at the proximal margin of the primary occlusion to facilitate clearing of the thrombus from the Indigo Aspiration Catheter tip. For the aspiration source, the Indigo Aspiration Catheter is used in conjunction with the Penumbra Aspiration Pump, which is connected using the Indigo Aspiration Tubing and the Indigo Pump/Canister Tubing. The Indigo Aspiration Catheter is provided with a steam shaping mandrel and rotating hemostasis valve, and a peelable sheath. The Indigo Separator is provided with an introducer and torque device. The devices are visible under fluoroscopy.

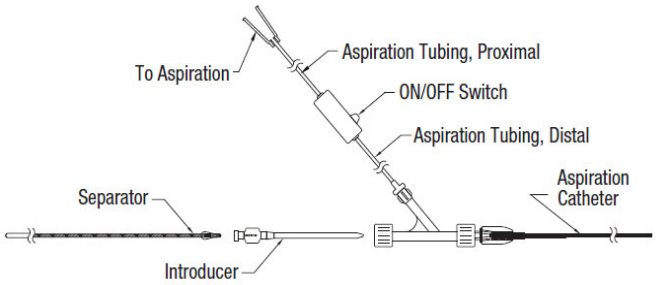
The Indigo Aspiration System received original US Food and Drug Administration (FDA) 510(k) clearance for the peripheral arterial indication in September 2012 under K121917. In May 2015 under K142870 the indication was expanded to the peripheral arterial and venous indication and is currently available in the United States. The Indigo Aspiration System received original CE mark on 18 September 2014 under Registration Number 252.943 and is currently available in Europe.

2.2 Indigo Aspiration Catheters and Separators

As part of the Indigo Aspiration System, the Indigo Aspiration Catheters and Separators are indicated for the removal of fresh, soft emboli and thrombi from vessels of the peripheral arterial and venous systems.

Indigo Aspiration Catheters and Separators are made of biocompatible materials commonly utilized for interventional devices. The Aspiration Catheter is provided sterile for single use and may be packaged in a kit with the Aspiration Tubing. The kit also contains a rotating hemostasis valve (RHV) and an introducer sheath.

Figure 1: Assembled Aspiration System with Indigo Separator



2.3 Indigo® Aspiration System, CAT RX Aspiration Catheter, and Separator™ 4

As part of the INDIGO Aspiration System, the INDIGO CAT RX Aspiration Catheters and INDIGO Separator 4 are indicated for the removal of fresh, soft emboli and thrombi from vessels in the coronary and peripheral vasculature.

General Description/Overview

The Indigo™ Aspiration System is comprised of several devices, which will be available based on applicable regulatory approvals for commercialization:

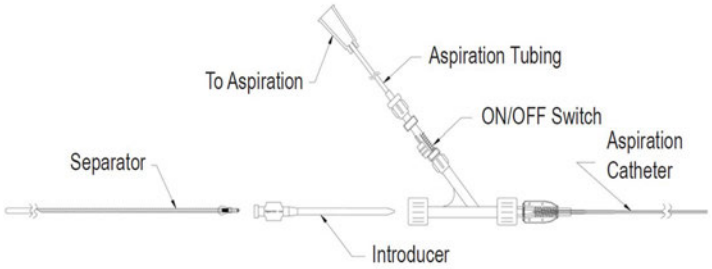
- Indigo CAT RX Aspiration Catheter
- Indigo Separator™ 4
- Indigo Aspiration Tubing
- Indigo Pump Canister/Tubing
- Penumbra Aspiration Pump

The Indigo Aspiration System is designed to remove thrombus from the vasculature using continuous aspiration. The Indigo CAT RX Aspiration Catheter is a dual lumen rapid exchange catheter that targets aspiration from the Pump directly to the thrombus, removing it via the Indigo Aspiration Tubing and depositing it in the Pump Canister. The Indigo Separator 4 may be used, if needed, to clear the lumen of the Indigo CAT RX Aspiration Catheter should it become blocked with thrombus. The Indigo CAT RX Aspiration Catheter is introduced through a guide catheter or long introducer sheath into the coronary or peripheral vasculature and guided over a guidewire to the site of the primary occlusion. The Indigo CAT RX Aspiration Catheter may be provided with a rotating hemostasis valve and a peelable sheath. The Indigo Separator 4 is provided with an introducer and torque device. The Indigo CAT RX Aspiration Catheter and Indigo Separator 4 are visible under fluoroscopy.

The Indigo CAT RX Aspiration Catheter, including the Separator 4 and the Aspiration Tubing, received US Food and Drug Administration (FDA) 510(k) clearance in May 2017 under K163618 and is currently available in the United States.

The Indigo CAT RX Aspiration Catheter, including the Separator 4 and the Aspiration Tubing may be used in other regions of the world as soon as applicable commercialization regulatory approvals are obtained.

Figure 2: Assembled Aspiration System with Indigo CAT RX and Separator™ 4



2.4 Aspiration Tubing

As part of the Indigo Aspiration System, the Indigo Sterile Aspiration Tubing is indicated to connect the Indigo Aspiration Catheters to the Penumbra Aspiration Pump.

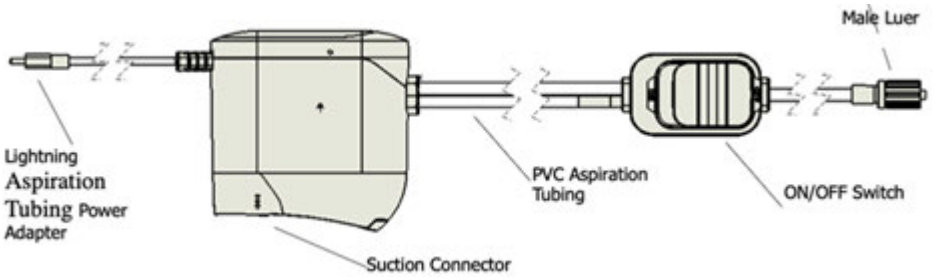
Lightning Aspiration Tubing is an additional component of the Indigo Aspiration System, which received original US Food and Drug Administration (FDA) 510(k) clearance in March 2020 under K193244 and is currently available in the United States.
The Lightning Aspiration Tubing may be used in other regions of the world as soon as applicable commercialization regulatory approvals are obtained.

Lightning Aspiration Tubing is designed to serve as a conduit to assist in thrombus removal and restoration of blood flow in the peripheral vasculature. Lightning facilitates the transfer of vacuum between the Aspiration Catheter and Penumbra Aspiration Pump while providing intermittent or continuous aspiration. Intended users for this device are physicians who have received appropriate training in surgical procedures and/or interventional techniques.

During scenarios when occlusion is detected (e.g. blood clot is engaged with an Indigo Aspiration Catheter), Lightning allows for continuous aspiration through the Indigo Aspiration Tubing. During scenarios when no occlusion is detected, Lightning is designed to minimize aspirated blood volume by converting to intermittent aspiration.

Consistent with the existing Indigo Aspiration Tubing, the operator/physician will be able to maintain manual control of flow via a flow control switch which will completely stop flow when in the “OFF” position.

Figure 2: Lightning Aspiration Tubing



2.5 Penumbra Aspiration Pump

The Penumbra Aspiration Pump is indicated as a vacuum source for Penumbra Aspiration Systems.

2.6 Penumbra Aspiration Pump Canister

A 1000 ml minimum canister with a stemmed lid is designed for use with the Pump. Each canister has a stop-flow filter to prevent excess fluid from entering the Pump Canister Tubing. The patient port on the lid is sized to accept the Suction Connector on the Aspiration Tubing. Graduations are placed on the canister in 50-ml and 100-ml increments. The canister lid is removable.

For the Pump MAX, Pump Canister Tubing is used to connect the Pump to the Pump Canister. This tubing has an inline filter to prevent fluid and contaminants from entering the pump. The tubing lumen remains open under full vacuum. The Penumbra ENGINE does not require this additional tubing. The Suction Connector is securely attached to the Pump Canister lid and Pump via press fit.

Further detailed description of all the devices listed in this protocol can be found within their respective Instructions For Use (IFU).

3 Risk Analysis

A thorough risk analysis was performed as part of design control requirements of the Quality System Regulation (21 CFR 820) and risk management activities were performed in accordance with ISO 14971

3.1 Risks Related to Aspiration Thrombectomy

It is anticipated that the treatment risks associated with the Indigo Aspiration System devices for removal of fresh, soft emboli and thrombi from vessels of the peripheral arterial and venous systems are unchanged since market introduction. In terms of the clinical risk, adverse events that may be associated with the use of the Indigo Aspiration System or with interventional procedures include, but may not be limited to:

- allergic reaction and anaphylaxis from contrast media
- acute occlusion
- air embolism
- arteriovenous fistula
- death
- device malfunction
- distal embolization
- emboli
- false aneurysm formation
- hematoma or hemorrhage at access site
- inability to completely remove thrombus
- infection
- hemorrhage
- ischemia
- kidney damage from contrast media
- neurological deficits including stroke
- vessel spasm, thrombosis, dissection, or perforation
- intimal disruption

- myocardial infarction
- emergent surgery
- fibrillation
- hypotension
- respiratory failure
- peripheral thromboembolic events

Risks that may be associated with the Lightning Aspiration Tubing include, but may not be limited to:

- allergic reaction and anaphylaxis from contrast media
- acute occlusion
- air embolism
- arrhythmia/fibrillation
- arteriovenous fistula
- death
- device malfunction
- distal embolization
- emergent surgery
- false aneurysm formation
- hematoma, hemorrhage, or blood loss at access site
- hematoma, hemorrhage, or blood loss
- hypotension
- inability to completely remove thrombus or control blood flow
- infection
- ischemia
- kidney damage from contrast media
- myocardial infarction
- neurological deficits including stroke
- respiratory failure
- thromboembolic events
- vascular complications (including vessel spasm, thrombosis, intimal disruption, dissection, or perforation)

Considerable testing has been completed to ensure that the Indigo Aspiration System does not pose a significant risk. Safety of the Indigo Aspiration System has been demonstrated through extensive pre-clinical testing in both bench-top and in vivo models. Critical design characteristics were evaluated to ensure that system components can withstand the rigor of clinical use. A thorough risk analysis was performed as part of design control recommendations of the Quality System Regulation (21 CFR 820). Risk management activities performed in accordance with ISO 14971did not identify any different risks or risk levels compared to other currently marketed devices.

A list of all anticipated adverse events is listed in the IFU for each device.

4 Study Overview

The intent of this study is to collect safety and performance data on Indigo Aspiration System in a patient population with LE ALI.

4.1 Study Design

STRIDE is a post-market, real world, prospective, single-arm, multi-center study looking at patients that present with LE ALI who are eligible for mechanical thrombectomy using the Indigo Aspiration System. Up to 130 subjects at up to 25 sites globally will participate.

4.2 Study Objectives/Endpoints

4.2.1 Primary Endpoints

- Target limb salvage rate at 1-month post-procedure

4.2.2 Secondary Endpoints

- Technical success defined as TIMI 2/3 flow rate immediate post-procedure
- Modified SVS runoff score immediate post-procedure as compared to baseline
- Improvement of Rutherford classification of one or more as compared to the pre-procedure
- Patency at 1 month
- Target limb salvage rate at 12 months post-procedure
- Rates of device related serious adverse events (SAEs)
- Major bleeding peri-procedure
- Mortality at 12 months

5 Study Population

5.1 Inclusion Criteria

1. Patient age ≥ 18
2. Patient presents with acute (≤14 days) occlusion of lower limb artery(ies) (below inguinal ligament)
3. Patient with a Rutherford Category I, IIa or IIb score
4. Frontline treatment with Indigo Aspiration System
5. Informed consent is obtained from either patient or legally authorized representative (LAR)

5.2 Exclusion Criteria

1. Life expectancy <1 year
2. Target vessel size <2 mm
3. LE ALI secondary to dissections, vasculitis and/or target vessel trauma
4. Amputation in the ipsilateral limb
5. Pregnancy or positive pregnancy test according to site specific standards of care (only required for women of childbearing potential, serum or urine acceptable)
6. Absolute contraindication to contrast administration
7. Patient is unwilling or unable to comply with protocol follow up schedule and/or based on the Investigator's judgment the patient is not a good study candidate

8. Currently participating in an investigational drug or device clinical trial that may confound the study endpoints. Patients in observational, natural history, and/or epidemiological studies not involving intervention are eligible.
9. Target thrombus in a saphenous vein bypass graft

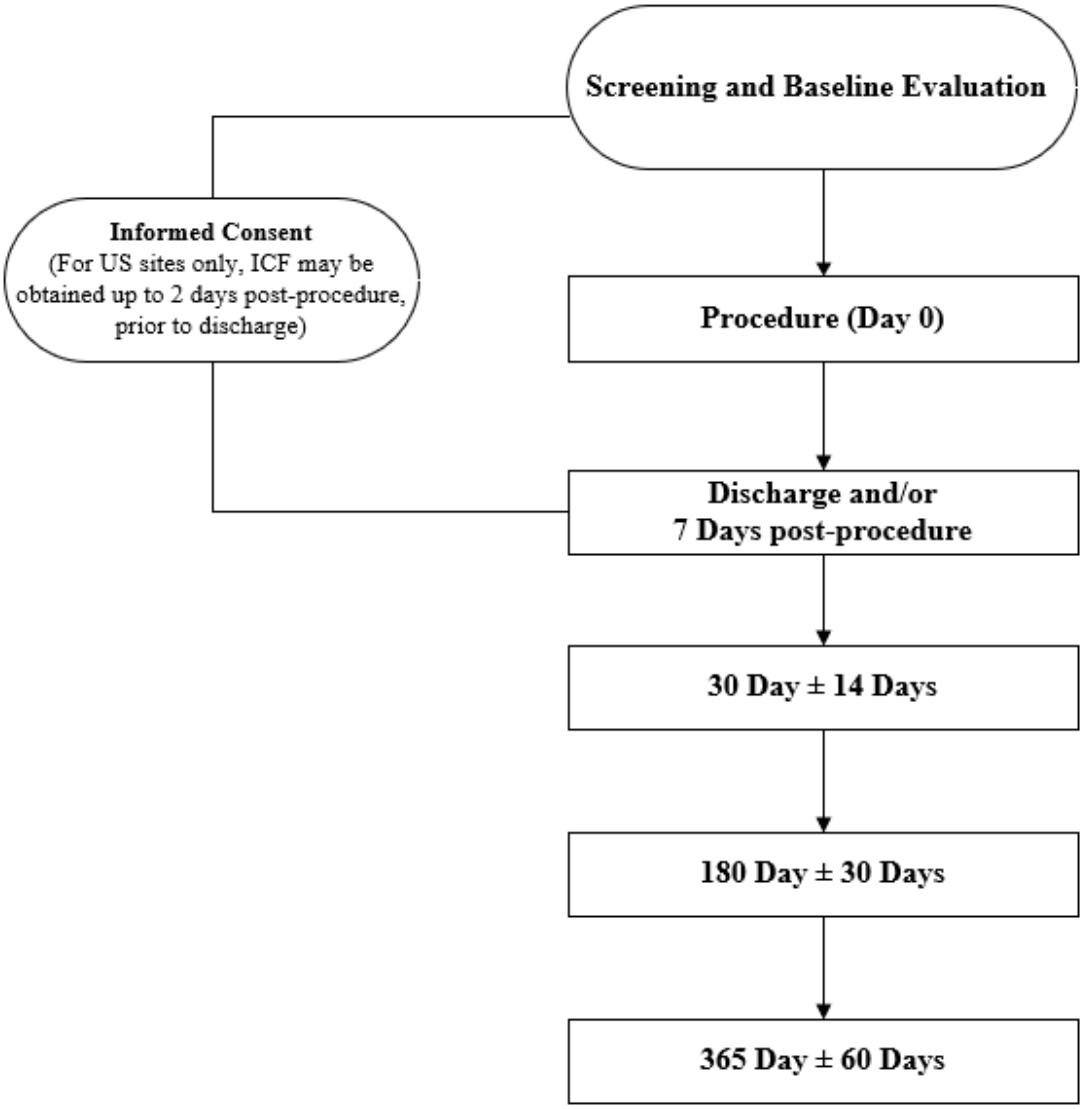
6 Study Procedures

6.1 Overview of Study Flow

Patients presenting with LE ALI should be assessed for eligibility. All sites will keep a screen failure log of all potential study candidates who are screened and not enrolled or screened, consented, and not enrolled. Reason(s) for exclusion will be recorded. Screening information, including screen failures, will be reported in Electronic Data Capture system (EDC).

Recruitment rates will be tracked over time for each site. The actual recruitment rates will be useful for planning further clinical studies and determining the widespread impact of the therapy.

Figure 1: Study Flow



6.2 Study Visits

Subjects enrolled in this study will follow the visit schedule below and will continue to receive routine practice/standard of care treatment at each follow up visit. Procedure is Day 0 for determining follow up visit dates.

1. Baseline Evaluation
2. Procedure (Day 0)
3. Discharge and/or 7 Days, whichever occurs first
4. 30 Day Follow-Up (± 14 days)
5. 180 Day Follow-Up (± 30 days)
6. 365 Day Follow-Up (± 60 days)

6.3 Recruitment and Screening

The target population are subjects ≥18 years of age who are presenting with LE ALI. No study specific screening tests or procedures are required for enrollment in the study. Standard of care evaluations will be used to confirm eligibility.

Potential study participants and/or the subjects' representative (e.g. LAR, next of kin, family member, etc.) will be identified by the study team at each site to obtain consent and determine eligibility.

6.4 Enrollment

Patients will be considered enrolled once informed consent is obtained per Institutional Review Board (IRB)/Ethics Committee (EC), the Indigo Aspiration Catheter has been inserted into the body as frontline treatment, and all eligibility is confirmed. Patients who fail to meet entry criteria will be considered a screen fail.

All follow-up visits will be conducted based on date of procedure, regardless of enrollment date.

6.5 Informed Consent

The Investigator or designee will obtain written informed consent from the subject or the subjects' representative (e.g. LAR, next of kin, etc.) using the current Institutional Review Board (IRB)/Ethics Committee (EC) approved consent form per IRB/EC policy.

For sites in North America, patients who have had a procedure (using the Indigo Aspiration System with the Indigo Aspiration Catheter as frontline) and all eligibility is confirmed, may be consented up to 2 calendar days post-procedure but prior to initial discharge, and enrolled in the study. The point of enrollment is defined as the date of informed consent or introduction of the Indigo Aspiration Catheter, whichever occurs later.

For sites in Europe, the informed consent process will be applied per local Ethics Committee approvals.

All informed consent documents used under this protocol will be consistent with applicable elements of Good Clinical Practice (ICH E6), ISO 14155,Clinical investigation

of medical devices for human subjects – Good Clinical Practice (GCP), FDA 21 CFR Part 50 Protection of Human Subjects, and will be approved by the site's reviewing IRB/EC prior to study initiation.

Any modification to the sample informed consent form made by the study site must be approved by the Sponsor and the IRB/EC before use. Each study site will provide the Sponsor with a copy of the IRB/EC approved consent forms. Informed consent completion will be monitored regularly by the Sponsor.

6.6 Baseline Evaluation

Subjects will be clinically evaluated in the same manner as any patient presenting with LE ALI. The medical history screen, available clinical exams obtained, and imaging information per institutional routine care will be evaluated to determine patient eligibility. Medication, standard of care lab values, and duplex ultrasound will be recorded.

Patients will be screened against study eligibility criteria during standard clinical practice. A signed study-specific IRB/EC-approved informed consent form (ICF) must be obtained from each potential subject (or the subjects' representative (e.g. LAR, next of kin, family member, etc.)) before performing any test that goes beyond standard clinical care.

These exams were chosen on the basis of their reliability, familiarity to the endovascular community, and comparability to other trials of LE ALI. All scores will be recorded in source documentation and entered into the electronic case report forms (eCRFs).

- Ankle-Brachial Index: ABI is a diagnostic test used to compare the systolic blood pressure at the ankle with the systolic blood pressure measured in the arm to gauge circulation.
- Rutherford Classification for LE ALI characterizes the acute presentation of patient clinical symptoms with objective findings, including arterial and venous Doppler signals (see APPENDIX II: Classifications and Scales).
- Vascular Quality of Life Questionnaire-6: VascuQoL-6 is a valid health-related quality of life questionnaire developed for use in peripheral arterial disease (see APPENDIX II: Classifications and Scales)

Baseline assessments will be performed, but not limited to the following:

1. Demographics
2. Confirm eligibility
3. General medical history
4. Pregnancy test (only required for women of childbearing potential)
5. Vital signs
6. Rutherford Classification
7. Ankle-Brachial Index (ABI)
8. Duplex Ultrasound (DUS), Computed Tomography Angiography (CTA), or Magnetic Resonance Angiography (MRA): will provide an initial diagnosis of LE ALI per institutional routine care
9. Medication review

10. VascuQoL-6
11. Laboratory Assessments as part of standard of care (SOC) (most recent results obtained within 30 days prior to index procedure are permitted as baseline data)
    a. INR, PT, aPTT/PTT, Platelets, WBC, RBC, Hgb, Hct, Creatinine, Glucose, BUN
    b. Additional SOC labs may be collected, if available

6.7 Procedure

All procedures will be conducted in accordance with standard of care guidelines for treatment of patients with LE ALI at each participating hospital. The procedure will occur after completion of baseline, clinical assessments, and pre-procedural imaging. The Indigo Aspiration System with the Indigo Aspiration Catheter must be used as frontline treatment.

6.7.1 Preparation for Treatment

All physicians will follow routine site practice guidelines to determine patient eligibility for the procedure.

6.7.2 Medication during Intervention

Any medical therapy necessary for patients with LE ALI should be used in accordance with the routine, standard of care guidelines at the study site.

6.7.3 Devices and Equipment

The follow devices/equipment will be used in addition to the standard devices used per site routine standard of care.

Please refer to the device description Section 2.0 for complete details.

Table 1: Devices that may be used during the procedure

Device Name	Description/Details
Indigo Aspiration Catheter	An intravascular aspiration thrombectomy catheter.
Indigo Separator™	Size matched accessory device for the Aspiration Catheter, intended to clear the distal end of the Aspiration Catheter lumen, should it be blocked with thrombus.
Indigo CAT RX Aspiration Catheter	A dual lumen rapid exchange intravascular aspiration thrombectomy catheter.
Indigo Separator™ 4	Size matched accessory device (optional) for the CAT RX Aspiration Catheter, intended to clear the distal end of the CAT RX Aspiration Catheter lumen, should it be blocked with thrombus.
Indigo Aspiration Tubing	Sterile, appropriately sized tubing that connects the Aspiration Catheter to the Penumbra Aspiration Pump. Lightning Aspiration Tubing facilitates the transfer of vacuum between the catheter and pump while providing intermittent or continuous aspiration.
Penumbra Aspiration Pump (Pump MAX and Penumbra ENGINE)	Provides continuous vacuum force
Indigo Pump Cannister and Tubing	The pump canister acts as a collection reservoir for materials aspirated by the Indigo Aspiration Catheter. For Pump MAX, pump tubing is used to connect the pump cannister to the aspiration pump. Penumbra ENGINE does not require this additional tubing.

6.7.4 Index Procedure

Subjects enrolled in the study should receive the SOC procedural care for LE ALI. At a minimum, the following information will be collected during Index Procedure, but not limited to:

1. Initial (pre-procedure)
    a. The following assessments will be administered before the Indigo Aspiration System and Aspiration Catheter is used:
        - Angiography: performed to assess procedure data points including the modified SVS runoff score.
        - Modified SVS runoff score: calculated using the angiogram by assessing the patency and degree of stenosis/occlusion in the popliteal artery and the three tibial vessels[17]
2. Target lesion characteristics
3. After Indigo Aspiration is complete (which can be considered Final if adjunctive procedures are not required) the following assessments will be administered, but not limited to:
    a. Angiography
    b. Modified SVS runoff score
    c. TASC II score, if applicable
4. Final (after all adjunctive procedures are complete)
    a. Angiography
    b. Modified SVS runoff score
5. Procedural details including key procedural timepoints
6. Adjunctive procedure(s)
7. Adverse event review
8. Medication review
9. Device deficiencies

Angiography should be uploaded per section 7.5 Image Upload.

6.7.5 Discharge and/or 7 days, whichever occurs first

Information will be collected during discharge and/or 7-day visit, (within 1 day prior to discharge or at day 7, whichever occurs first), but not limited to the following:

1. Vital signs
2. Rutherford classification
3. ABI
4. Adverse event review

In addition to the assessments above, the following will be collected at discharge:

1. The location that the subject is discharged to and the date of discharge
2. If any adverse events occurred, it will be reported on the Adverse Event Form

3. Details of each hospital unit that the subject was admitted to during the index admission will be recorded
4. Medication use

6.7.6 30 Day Follow Up Visit (± 14 days)

Information will be collected during the 30-day follow up visit, but not limited to the following:

1. Vital signs
2. Rutherford classification
3. DUS, CTA, or MRA
4. ABI
5. VascuQoL-6
6. Medication review
7. Adverse event review

6.7.7 180 Day Follow Up Visit (± 30 days)

Information will be collected during the 180-day follow up visit, but not limited to the following:

1. Vital signs
2. Rutherford classification
3. DUS, CTA or MRA
4. ABI
5. VascuQoL-6
6. Medication review
7. Adverse event review

6.7.8 365 Day Follow Up Visit (± 60 days)

Information will be collected during the 365-day follow up visit, but not limited to the following:

1. Vital signs
2. Rutherford classification
3. DUS, CTA, or MRA
4. ABI
5. VascuQoL-6
6. Medication review
7. Adverse event review

Table 2: Schedule of Assessments

ACTIVITY	Screening and Baseline Evaluation	Procedure (Day 0)	Discharge and/or 7 Days (whichever occurs first)	30 Day (± 14 days)	180 Day (± 30 days)	365 Day (± 60 days)
Written Informed Consent	X[1]					
Inclusion/Exclusion	X					
Medical History	X					
Pregnancy Test	X[2]					
Vital Signs	X		X	X	X	X
Rutherford Classification	X		X	X	X	X
Ankle-Brachial Index	X		X	X	X	X
DUS, CTA, or MRA	X			X	X	X
VascuQoL-6	X			X	X	X
Laboratory Assessments	X	X[3]				
Modified SVS Runoff Score and TASC II Score		X[4,6]				
Angiography		X[4,5]				
Procedure		X				
Adverse Event Review		X	X	X	X	X
Medication Review	X	X	X	X	X	X

[1] For US only, patients who have had an index procedure with the Indigo® Aspiration System and Aspiration Catheter used frontline, prior to informed consent, may be consented up to 2 calendar days post procedure but prior to discharge
[2] Only required for women of childbearing potential
[3] Laboratory assessments performed as part of standard of care
[4] Modified SVS Runoff Score will be required at multiple timepoints within the visit
[5] Angiography images must be uploaded to image management system for core lab review
[6] Modified SVS Runoff score is required for all subjects. TASC II score is only required when lesions are in the aortoiliac and/or femoropopliteal segments

7 Investigator Responsibilities

7.1 Institutional Review Board / Ethics Committee Approval

Prior to enrolling patients into the study, the Investigator will ensure that proper IRB/ EC approval is obtained in accordance with applicable local laws and regulations. The IRB/EC shall approve all study documents as appropriate, including but not limited to the final protocol, amendments to the protocol, IFU (where applicable), and the informed consent, per FDA 21 CFR part 56; Institutional Review Boards or applicable EC requirements. The investigator should keep the IRB/EC informed of any serious adverse event, or serious device defects as per their policy.

The Investigator will report to the Sponsor or designee immediately if the approval to conduct the investigation is withdrawn by the IRB/EC or Competent Authority as required. The report will include a complete description of the reason(s) for which approval was withdrawn.

7.2 Informed Consent

The Investigator is responsible for ensuring that completed informed consent is obtained in accordance with Section 6.5 of this protocol and according to country and local requirements prior to conducting any study-related assessments prior to enrollment of patients in the study.

7.3 Adherence to Protocol/Amendments and Applicable Law

The Investigator is responsible for overseeing, ensuring that the study is conducted, and completing the study according to this protocol and in accordance with the relevant aspects of ISO 14155, 21 CFR 50, 54, and 56, Declaration of Helsinki, along with any conditions imposed by the reviewing IRB/EC, and all other applicable regulations. The Investigator shall approve and adhere to this protocol and any amendments that arise during the course of the study. The Investigator has overall responsibility of the conduct of this study including oversight of all delegated individuals, tasks and all activities conducted under this protocol pursuant to FDA 21 CFR part 820 and Investigator responsibilities.

It is the Investigator’s responsibility to ensure that the Institution’s staff assisting with the study have the appropriate qualifications, are fully instructed on the study procedures, and will respect study confidentiality.

7.4 Case Report Form Completion

The Investigator and study staff shall complete the eCRFs associated with this study. Subject numbers shall be used to identify individual participants in this study. The eCRFs should be a complete and accurate record of subject data collected during the study according to relevant aspects of ISO 14155, FDA 21 CFR 11, Electronic Records; Electronic Signatures and GCP requirements. It is the Investigator's responsibility to ensure the quality of the data collected and recorded is appropriate and collected in accordance with GCP and all applicable regulations. Data entry will be performed by the investigational site(s). Investigators are responsible for completion and timely submission

of data to Penumbra, Inc. Every reasonable effort should be made to complete data entry within 7 business days of data collection.

7.5 Image Upload

Sites will be provided with instructions for how image timepoints should be collected and submitted within 14 days of the acquisition of the required imaging. If the site is unable to provide the images within this time frame, the appropriate Sponsor contact should be notified.

Angiography will be uploaded to an image management system for Core Lab review. Instructions for image collection and upload will be provided. Study staff shall ensure that no images contain any personally identifying information about the subject or study site (e.g. Physician name, Institution name, patient name, etc.).

7.6 Reporting

The Investigator will be responsible for reporting the following:

7.6.1 Adverse Events

Adverse events (AE) must be recorded by the Investigator on the eCRFs and will be monitored during the entire study. All SAEs and procedure and device related AEs will be collected starting at Day 0 through final follow up for enrolled subjects. Additionally, bleeding complications should be reported.

Minimum requirements of data to be recorded are: event term, event start date, seriousness, action taken, outcome and device-relatedness or causality.

In order to ensure prompt reporting of AEs, all reportable AEs (as well as all related study data) are required to be entered into the EDC in a timely manner. Any suspected Unanticipated Adverse Device Events (UADEs)/ Unanticipated Serious Adverse Device Effects (USADE) should be reported immediately by calling the Sponsor. All device related Serious Adverse Events (SAEs) should be reported in the EDC within 72 hours of the site staff first being made aware of the occurrence of the SAE. If the EDC is unavailable, an email can be sent to Penumbra.

The Investigator must report adverse events to the IRB/EC according to local requirements. The Investigator is responsible for reporting time frames and complying with local or national requirements. In addition, the Investigator will report to the sponsor and IRB/EC any device malfunctions that could have led to a SAE, if required by national regulations or by local authorities.

For the purpose of reporting within this protocol, pre-existing conditions or planned procedures for pre-existing conditions are not reportable as AEs unless there is worsening of the condition with an increase in severity or frequency during the course of the study. All deaths will be reported regardless of causality. When reporting a death, the primary condition or diagnosis that contributed to the fatal outcome should be reported as a SAE with an outcome of death. Only a single cause

of death should be reported in EDC. If the cause of death is unknown, report "unknown cause of death" as a SAE.

7.6.2 Analysis of Adverse Events

A Sponsor Medical Monitor will review the pre-defined categories of adverse events as they are reported throughout the duration of the study. The Sponsor Medical Monitor will collect redacted source documents and review adverse events.

7.6.2.1 Definitions

- **Adverse Event (AE)**: Any undesirable clinical event occurring in a patient during a clinical trial, whether or not it is considered related to the study device or whether anticipated or unanticipated. This includes a change in a patient's condition or laboratory results that has or could have a deleterious effect on the patient's health or well-being or untoward clinical sign in users or other persons related to use of medical device.

- **Adverse Device Effect (ADE):** An adverse event related to the use of the study medical device.

- **Definition of SAE**
  A SAE is an event that:
  - Led to death
  - Led to a serious deterioration in the health of the patient that:
    - Resulted in life-threatening illness or injury
    - Resulted in chronic disease
    - Resulted in permanent impairment of a body structure or a body function
    - Required in-patient hospitalization or prolongation of existing hospitalization
    - Resulted in medical or surgical intervention to arrest permanent impairment to body structure or a body function
    - Led to fetal distress, fetal death or a congenital abnormality or birth defect

- **Unanticipated Adverse Device Event (UADE):** An unanticipated adverse device effect is any serious adverse effect on health or safety or any life-threatening problem or death caused by, or associated with, a device, if that effect, problem, or death was not previously identified in nature, severity, or degree of incidence in the current risk assessment.

7.6.2.2 Relationship to the Study Device

An AE is considered to be device-related when it is reasonable to believe that the event may have been caused by or is related to the device. The following

definitions will be used to assess the relationship of the adverse event to the use of study device.

- **Definite:** The temporal sequence is relevant and the event abates upon device application completion/removal, or reappearance of the event on repeat device application

- **Probable:** The temporal sequence is relevant or the AE abates upon device application completion/removal or the AE cannot be reasonably explained by the subject's condition or comorbidities. The AE is related or most likely associated with the device

- **Possible:** The temporal sequence between the device and the AE is such that the relationship is not unlikely or there is no contradicting evidence that can reasonably explain the subject's condition. There is a possibility of a relationship between the AE and the device

- **Unrelated:** The AE is not associated with the device. There is no relation between the AE and the device

Similar grading will be used for assessing the relationship to index procedure and to index LE ALI.

7.6.3 Device Deficiencies

All device deficiencies related to the identity, quality, durability, reliability, safety or performance of the study device shall be documented and reported through the standard commercial process and within the EDC. Investigators must report all possible device deficiencies associated with the device observed during the study. This includes unexpected outcomes or device deficiencies that might have led to a serious adverse event if a) suitable action had not been taken or b) intervention has not be made or c) if circumstances had been less fortunate.

Device manufacturers are required to report qualifying medical device incidents to the relevant national competent authorities. An incident is defined as "any malfunction or deterioration in the characteristics and/or performance of a device, as well as any inadequacy in the labeling or the instructions for use which, directly or indirectly, might lead to or might have led to the death of a subject or to a serious deterioration in their state of health." A deterioration in state of health is not considered unanticipated if the condition leading to the event was considered in a risk analysis.

7.6.4 Protocol Deviation

Deviations defined in this protocol should be clearly documented, if identified during monitoring or through other means. For this study, deviations should be reported for the following categories:

- Inclusion/exclusion criteria deviation(s)
- Informed consent deviation(s)

7.7 Records Retention

The Investigator shall maintain the records associated with this study for a period as defined by local regulations A Trial Master File (TMF) will be used as the master repository for all site and Sponsor regulatory documents. These records include the following:

- Correspondence with the Sponsor or designee, the Medical Monitor, and other investigators
- Subject source records, including but not limited to: Informed Consent Forms, copies of all completed eCRFs, and supporting documents (laboratory reports and reports of diagnostic tests, medical records, etc.)
- All versions of the study protocol
- Documentation of protocol deviations
- Reports of any serious adverse event and serious adverse device effects, if any
- A copy of all approvals related to the clinical study
- The approved, blank, informed consent form
- All approval/acknowledgment letters from the IRB/EC for all versions of the study protocol, ICF and other documents
- Clinical Trial Agreement(s)
- Signed and dated curriculum vitae for all study personnel
- Medical licenses or equivalent for the Principal Investigator and all participating sub-Investigators
- Financial disclosure for the Principal Investigator and all participating sub-Investigators
- All required regulatory documents such as Delegation of Authority and training logs
- Signed Protocol Signature Page(s)
- Data Privacy Agreements for the Principal Investigator and all hospital staff directly involved in the study (EU sites only)
- Product liability certificates (EU sites only)

8 Sponsor Responsibilities

8.1 Training

The Sponsor is responsible for providing training on the protocol, study device, eCRF completion, image upload as applicable for all study staff per delegation of authority log.

8.2 Investigator List

The Sponsor shall keep a list of the names and addresses of the clinical investigators for the study, including the investigator role and his specialty.

8.3 Adverse Event Reporting

The Sponsor shall evaluate adverse event reports received from the study sites and found during data monitoring and shall report them to the appropriate regulatory bodies and other study sites as necessary.

8.4 Data Monitoring

Penumbra is responsible for ensuring that the study is conducted according to applicable portions of the regulations (US Food and Drug Administration 21 CFR 812, ISO 14155). A Penumbra employee or designate will conduct the following site visits:

8.4.1 Site Qualification Visit

Conducted to ensure the study site has the appropriate staff, facilities, and expertise to participate in the study. Site Qualification can be waived under certain circumstances.

8.4.2 Site Initiation Visit

Conducted to train the study staff on use of the device, study requirements, and other relevant training.

8.4.3 Interim Monitoring Visit

Conducted as needed to ensure the study site is operating in compliance with this protocol, continues to have the appropriate staff and facilities, and is correctly completing the eCRFs.

To ensure that Investigators and their staff understand and accept their defined responsibilities, the Sponsor will maintain regular correspondence and perform periodic site visits during the course of the study to verify the continued acceptability of the facilities, compliance with the protocol, and maintenance of complete records. Clinical monitoring will include review and resolution of missing or inconsistent data and source document checks to ensure the accuracy of the reported data. Informed consent, eCRFs and medical records for all enrolled and screen failed subjects will be made available to the Sponsor for review and collection.

8.4.4 Site Close Out Visit

Conducted to ensure all study and regulatory-related activities have been completed prior to site closure.

8.5 Data Management

eCRFs will be used at all study sites. All study data will be entered into commercially available web-based EDC. Data entry will be performed by the study site personnel. Investigators are responsible for completion and timely submission of the data to the Sponsor. Every reasonable effort should be made to complete data entry within 7 days of data collection. This EDC system requires no on-site software installation or specific

hardware to operate. Investigators, clinical coordinators, data managers, and Penumbra clinical personnel access project information and study data centrally via a web browser.

Automated data quality checks will display warnings for invalid data. Additionally, manual review of data listings may be used to identify data discrepancies or inconsistencies. The study site may be queried for clarification concerning eCRF discrepancies or inconsistencies identified. If eCRF corrections are necessary, they will be made by the Investigator or an authorized member of the Investigator's staff that is delegated to eCRF/EDC. Questions or problems with submitted data will be addressed with the Principal Investigator via an electronic querying system, or through direct contact. The Investigator will review the eCRFs for completeness and accuracy and provide his/her electronic signature and date to eCRFs as evidence thereof. Any data items that have been changed will require reapplication of the electronic signature.

Study personnel will have individual login and password to access the clinical study information based upon each individual's roles and responsibilities. The application provides hierarchical user permission data entry, viewing, and reporting options.

All data entry and data update information, including the date and person performing the action, will be available via the audit trail, which is part of the EDC system.

All eCRFs and data files will be secured to ensure confidentiality. Investigators are required to maintain source documents required by the protocol, including laboratory results, reports, supporting medical records, and Informed Consent Forms. The source documents will be used during the regular monitoring visits to verify information entered on the eCRFs.

9 Ethical Requirements

9.1 Declaration of Helsinki

The study will be performed in accordance with the applicable aspects of ISO 14155, recommendations guiding physicians in biomedical research involving human subjects adopted by the 18th World Medical Assembly, Helsinki, Finland (1964 and later revisions), ICH and US FDA GCP guidelines.

It is the responsibility of the Investigator to obtain approval of the study protocol from the IRB/EC and to keep the IRB/EC informed of any serious adverse event, serious adverse device effects, and amendments to the protocol. All correspondence with the IRB/EC should be filed by the Investigator and copies sent to the Sponsor or its designee.

9.2 Informed Consent

The Investigator is responsible for ensuring that a completed informed consent is obtained in accordance with Section 6.5 of this protocol, as delegated by the site-specific Delegation of Authority, and according to country and local requirements.

9.3 Subject Data Protection

Each subject will be assigned a unique subject identification number at the time of enrollment. This subject identification number will be retained throughout the study. Study sites will keep a log that notes the subject's name and corresponding subject identification number. All eCRFs will be tracked, evaluated, and stored using only the subject ID number. No personal identifying information will be included on the eCRFs.

The informed consent form will notify subjects that study monitors, Sponsor's representatives, auditors, and representatives of government agencies and ethics committees will have access to personal identifying information to ensure that data reported on the eCRFs corresponds to the person who signed the consent form and the information contained in the source documentation.

Each subject must be informed that the data collected will be stored and analyzed by computer and the applicable national regulations for handling of computerized data will be followed. Furthermore, each subject should be informed about the possibility of inspection of relevant parts of the hospital records by the Sponsor, or other Health Authorities, including the FDA; and that the data will be electronically stored in the United States.

10 Statistical Procedures

10.1 General Statistical Considerations

All confidence intervals presented will be two-sided. All statistical tests will be two-tailed with a significance level of 0.05. Descriptive statistics will be provided. This includes the number of observations, mean, median, standard deviation, inter-quartile range, minimum and maximum for continuous variables and counts and percentages for discrete variables. Analyses will be conducted using SAS (SAS Institute, Cary, NC). The specific details of the planned analyses are described completely in the statistical analysis plan.

10.2 Sample Size Estimation for the Primary Outcome

The sample size calculations assume that 94% (108/115) of the study subjects achieve target limb salvage 1-month post-procedure as compared to 95.7% patients undergoing current standard of care treatments. Based on a binomial analysis with a non-inferiority margin of 10%, a study of 115 enrolled subjects will have 80% power with a one-sided alpha of 0.025. The sample size was adjusted to 130 subjects to account for approximately 10% attrition.

10.3 Control of Systematic Error and Bias

The study will be conducted under a common protocol for each study site with the intention of pooling the data for analysis. Every effort will be made to promote consistency in study execution at each study site.

10.4 Missing Data and Imputation Methods

Every effort will be made to keep all missing data, particularly at the 30-day follow-up visit, to a minimum. Some data may be missing, mainly due to lost-to-follow-up (LTFU)

subjects. The primary analysis will be data as observed. Sensitivity analysis will be performed.

10.5 Definition of Populations

10.5.1 Screened

Screened subjects are all subjects considered for participation in the study, whether or not they sign informed consent.

10.5.2 Screen Failure

Screen failure subjects are all subjects considered for participation in the study, who failed to meet inclusion criteria or met exclusion criteria. Patients can be screen failed based on general criteria. These patients may or may not have signed an informed consent form.

10.5.3 Enrolled

An eligible patient is considered enrolled once informed consent is obtained and Indigo Aspiration System has been inserted into the body.

10.5.4 Completed

Completed subjects are all subjects who were enrolled and completed the study follow-up or were known to have died prior to the follow-up timepoint are considered completed. The completed subject metric will be provided for 365-day follow-up.

10.5.5 Early Termination

Early termination subjects are all subjects who were enrolled but did not complete follow-up and were not known to have died are considered early termination subjects. The early termination subject metric will be provided for 365-day follow-up.

10.6 Definition of Analysis Populations

10.6.1 Intent to Treat Sample

As the primary analysis, all performance and safety outcome measures will be analyzed under the intent-to-treat (ITT) principle. Under this principle, the ITT sample includes all subjects who are enrolled. This population is the primary analysis population.

10.7 Interim Analysis

No interim analyses are planned for the purpose of terminating the study for a positive result. Interim analysis without hypothesis testing may be performed for regulatory submission or publication of study results. No adjustments will be made to the confidence bounds for the final analysis.

10.8 Statistical Analysis of Primary Endpoint

The primary endpoint is the target limb salvage rate at 1-month post-procedure. The primary safety analysis will be the difference between the study group and the combined historical control rate of 95.7% observed under current standards of care.

The null hypothesis is that the difference between the primary endpoint rate and the standard of care at 1-month post-procedure is less than or equal to -10%. The alternative hypothesis is the difference at 1-month post-procedure is greater than -10%. Formally, the null and alternative hypotheses to be tested are as follows:

H_0: $P_{study} - 0.957 \leq -0.10$

H_A: $P_{study} - 0.957 > -0.10$

Where P_{study} is the proportion of patients who achieve target limb salvage 1-month post-procedure.

The primary safety endpoint is met if the lower limit of the 95% confidence interval of the primary endpoint rate is greater than 85.7%. The primary safety analysis will be unadjusted.

10.9 Secondary Statistical Analysis

The secondary performance and safety endpoints will be assessed via proportions based on the endpoint criteria and 95% confidence intervals will be presented. The modified SVS runoff score will be analyzed both as a categorical and a continuous outcome. Survival estimates will also be utilized to evaluate the time-to-event using Kaplan-Meier methodology for events through 365 days. With the date of procedure set at day 0, any event occurring on or before day 365 will be included.

10.10 Analysis of Adverse Events

All adverse events will be summarized by showing the number and percent of subjects which report the event. Events will also be reported by relationship to the procedure or device. Adverse events judged as probably or definitely related to the Indigo Aspiration System will be analyzed as device-related.

10.11 Baseline Characteristics

Baseline data including, but not limited to demographics, clinical characteristics, and angiographic characteristics will be summarized using descriptive statistics.

10.12 Pooling Across Centers

Analyses will be presented by treatment group using data pooled across sites. Adjusted analysis using key baseline variables and the study site will be used to assess any potential site effects. This analysis will be performed on the intent-to-treat population.

10.13 Final Report

A final report will be completed, even if the study is prematurely terminated. At the conclusion of the study, a multi-center abstract reporting the results will be prepared and may be presented at a major meeting(s). A multi-center publication may also be prepared for publication in a reputable scientific journal. The publication of results from any single center experience within the study is not allowed until the aggregate study results have been published, unless there is written consent from the Sponsor.

11 Study Committees and Core Labs

11.1 Imaging Core Lab

The Imaging Core Lab is composed of independent medical doctor(s) who are not participants in the study. The Imaging Core Lab is responsible for assisting in the development of specific criteria used for the categorization of clinical endpoints in the study. A web-based electronic database will be provided for the Imaging Core Lab to review and adjudicate images. Additional details related to the core lab are specified in the Imaging Core Lab Charter

The independent imaging core lab will review images from the pre-procedure and post-procedure (pseudonymized) angiograms to determine at minimum, the TIMI scores. An Imaging core lab charter will provide procedures for core lab review.

12 Study Administration

12.1 Stopping the Study Based on Interim Safety Data

The Medical Monitor will review periodic safety reports of all AEs and SAEs. The study can be temporarily suspended to evaluate any negative safety trends.

12.2 Clinical Study Termination/Withdrawal

Subjects may be terminated or withdrawn from the study for the following reasons:

- Voluntary withdrawal of consent— meaning that a subject voluntarily chooses not to participate further in the study. All data collected up to the withdrawal of consent will be maintained in the study database. Withdrawn subjects will not have any additional follow-up and will not be replaced.
- Lost to follow-up — a subject will be considered lost-to-follow-up when contact is not achieved at the last required follow-up visit window. At a minimum, the effort to obtain follow-up information will include three (3) attempts to make contact via telephone or e-mail and if unsuccessful, then a letter from the Investigator sent via courier or other traceable method will be sent to the subject's last known address. These efforts to obtain follow-up will be recorded in the subject's study files.
- Subjects may also be withdrawn at the Investigator's discretion if within their best interest. A subject's participation in the clinical study will be terminated if the Investigator believes that this is in the subject's best medical interest or if the subject no longer complies with the clinical study requirements.

The sponsor may temporarily suspend or prematurely terminate the study at any time for the following reasons:

- Suspicion of risk to subjects
- If no positive IRB/EC decision is obtained or if the judgement of the IRB/EC is revoked
- If the applicable regulatory body has made an irrevocable objection
- If it transpires that continuation of study cannot serve any scientific purpose, and this is confirmed by the IRB/EC
- Business reasons

The Sponsor will document reasons for study suspension or premature termination and notify the PIs. The Sponsor will ensure that the IRB/ECs and regulatory authorities are notified in a timely manner.

The Sponsor will continue to provide resources to fulfil the obligations from the study protocol and existing agreements for following up the subjects enrolled in the study.

The Principal Investigators will promptly inform the enrolled subjects at his/her site, if appropriate.

If the Sponsor temporarily suspends the study and wishes to resume it, the Sponsor will inform the PIs, IRB/ECs, and (if appropriate) regulatory authorities. The Sponsor will provide a rationale for resuming the study. IRB/ECs must provide written approval before the study is resumed.

12.3 Missing Visits

Every effort should be made to bring subject in to scheduled follow-up visits. Any study subject who does not attend a scheduled follow-up visit should be contacted by site personnel to reschedule. If the missed visit was due to an adverse event, an AE eCRF must be completed and any reporting requirements met.

12.4 Protocol Adherence and Amendments

Prior to beginning the study, the Principal Investigator must sign the protocol signature page documenting his/her agreement to conduct the study in accordance with this protocol. Deviations outlined in section 7.6.4 must be documented and reported to Penumbra as soon as possible, and to the IRB/EC per local guidelines and government regulations.

12.5 Study Registration

The study will be registered in a publicly accessible trial study database (e.g., clinical trials.gov) prior to study initiation.

13 Publication of Information

All information and data generated in association with this study will be held in strict confidence and remain the sole property of the Sponsor. The Investigator agrees to use this

information for the sole purpose of completing this study and for no other purpose without written consent from the Sponsor.

The results of this study may be offered for publication. The Investigators and the Sponsor shall collaborate in the writing of the study to ensure accuracy. All information not previously published concerning the test device and research, including patent applications, manufacturing processes, basic scientific data, etc., is considered confidential and should remain the sole property of Penumbra. The Investigator agrees to use this information only in connection with this study and will not use it for other purposes without written permission from the Sponsor.

14 Contact Information

The address of Penumbra Incorporated, who is funding this research:

Penumbra, Inc.
One Penumbra Place
Alameda, CA 94502
Tel. (510) 748-3200
Fax (510) 814-8305

Key contacts for the Study include:

15 References

1. Gilliland, C., Shah, J., Martin, J.G., and Miller, M.J., Jr. (2017). Acute limb ischemia. *Tech Vasc Interv Radiol*. 20(4): p. 274-280.
2. Gerhard-Herman, M.D., Gornik, H.L., Barrett, C., Barshes, N.R., Corriere, M.A., Drachman, D.E., . . . Walsh, M.E. (2017). 2016 aha/acc guideline on the management of patients with lower extremity peripheral artery disease: A report of the american college of cardiology/american heart association task force on clinical practice guidelines. *J Am Coll Cardiol*. 69(11): p. e71-e126.
3. Aboyans, V., Ricco, J.B., Bartelink, M.E.L., Bjorck, M., Brodmann, M., Cohnert, T., . . . Zamorano, J.L. (2018). Editor's choice - 2017 esc guidelines on the diagnosis and treatment of peripheral arterial diseases, in collaboration with the european society for vascular surgery (esvs). *Eur J Vasc Endovasc Surg*. 55(3): p. 305-368.
4. Howard, D.P., Banerjee, A., Fairhead, J.F., Hands, L., Silver, L.E., and Rothwell, P.M. (2015). Population-based study of incidence, risk factors, outcome, and prognosis of ischemic peripheral arterial events: Implications for prevention. *Circulation*. 132(19): p. 1805-15.
5. Baril, D.T., Ghosh, K., and Rosen, A.B. (2014). Trends in the incidence, treatment, and outcomes of acute lower extremity ischemia in the united states medicare population. *J Vasc Surg*. 60(3): p. 669-77 e2.
6. Acar, R.D., Sahin, M., and Kirma, C. (2013). One of the most urgent vascular circumstances: Acute limb ischemia. *SAGE Open Med*. 1: p. 2050312113516110.
7. Creager, M.A., Kaufman, J.A., and Conte, M.S. (2012). Clinical practice. Acute limb ischemia. *N Engl J Med*. 366(23): p. 2198-206.
8. Rutherford, R.B., Baker, J.D., Ernst, C., Johnston, K.W., Porter, J.M., Ahn, S., and Jones, D.N. (1997). Recommended standards for reports dealing with lower extremity ischemia: Revised version. *J Vasc Surg*. 26(3): p. 517-38.
9. Leung, D.A., Blitz, L.R., Nelson, T., Amin, A., Soukas, P.A., Nanjundappa, A., . . . Simoni, E.J. (2015). Rheolytic pharmacomechanical thrombectomy for the management of acute limb ischemia: Results from the pearl registry. *J Endovasc Ther*. 22(4): p. 546-57.
10. Investigators, T.S. (1994). Results of a prospective randomized trial evaluating surgery versus thrombolysis for ischemia of the lower extremity. The stile trial. *Ann Surg*. 220(3): p. 251-66; discussion 266-8.
11. Ouriel, K., Shortell, C.K., DeWeese, J.A., Green, R.M., Francis, C.W., Azodo, M.V., . . . Marder, V.J. (1994). A comparison of thrombolytic therapy with operative revascularization in the initial treatment of acute peripheral arterial ischemia. *J Vasc Surg*. 19(6): p. 1021-30.
12. Ouriel, K., Veith, F.J., and Sasahara, A.A. (1996). Thrombolysis or peripheral arterial surgery: Phase i results. Topas investigators. *J Vasc Surg*. 23(1): p. 64-73; discussion 74-5.
13. Morrison, H.L. (2006). Catheter-directed thrombolysis for acute limb ischemia. *Semin Intervent Radiol*. 23(3): p. 258-69.
14. Shishehbor, M.H. (2014). Acute and critical limb ischemia: When time is limb. *Cleve Clin J Med*. 81(4): p. 209-16.

15. Kwok, C.H.R., Fleming, S., Chan, K.K.C., Tibballs, J., Samuelson, S., Ferguson, J., . . . Jansen, S.J. (2018). Aspiration thrombectomy versus conventional catheter-directed thrombolysis as first-line treatment for noniatrogenic acute lower limb ischemia. *J Vasc Interv Radiol*. 29(5): p. 607-613.
16. Saxon, R.R., Benenati, J.F., Teigen, C., Adams, G.L., Sewall, L.E., and Trialists, P. (2018). Utility of a power aspiration-based extraction technique as an initial and secondary approach in the treatment of peripheral arterial thromboembolism: Results of the multicenter prism trial. *J Vasc Interv Radiol*. 29(1): p. 92-100.
17. Duval, S., Keo, H.H., Oldenburg, N.C., Baumgartner, I., Jaff, M.R., Peacock, J.M., . . . Hirsch, A.T. (2014). The impact of prolonged lower limb ischemia on amputation, mortality, and functional status: The friends registry. *Am Heart J*. 168(4): p. 577-87.
18. Hardman, R.L., Jazaeri, O., Yi, J., Smith, M., and Gupta, R. (2014). Overview of classification systems in peripheral artery disease. *Semin Intervent Radiol*. 31(4): p. 378-88.
19. Surgery, T.W.G.a.t.S.f.V. (2000). Clinical classification of acute limb ischemia. *J of Vasc Surg.* 31(1): p. S142-S145.
20. Stoner, M.C., Calligaro, K.D., Chaer, R.A., Dietzek, A.M., Farber, A., Guzman, R.J., . . . Society for Vascular, S. (2016). Reporting standards of the society for vascular surgery for endovascular treatment of chronic lower extremity peripheral artery disease. *J Vasc Surg*. 64(1): p. e1-e21.
21. Sarkar, A., Grigg, W.S., and Lee, J.J., Timi grade flow, in Statpearls. 2020: Treasure Island (FL).
22. Nordanstig, J., Wann-Hansson, C., Karlsson, J., Lundstrom, M., Pettersson, M., and Morgan, M.B. (2014). Vascular quality of life questionnaire-6 facilitates health-related quality of life assessment in peripheral arterial disease. *J Vasc Surg*. 59(3): p. 700-7.

16 APPENDIX I: Definitions and Classifications

16.1 Definitions

Acute Limb Ischemia:	Acute (≤14 days), severe hypoperfusion of the limb characterized by these features: pain, pallor, pulselessness, poikilothermia (cold), paresthesias, and paralysis.[2]
Amputation:	The complete or partial removal of lower limb or extremity. Minor amputation is surgical removal of a portion or segment of the lower extremity distal or through the tarsometatarsal joints. Major amputation is complete or partial removal that occurs proximal to the tarsometatarsal joints.[22]
Limb Salvage:	Condition of extremity with potential to secure viability and preserve motor function to the weight-bearing portion of the foot if treated.[2] Limb is considered salvagable if amputation is performed at or distal to tarsal/metatarsal joint.[22]
Major Bleeding:	Fatal or leading to a drop in hemoglobin of ≥5 g/dl, or significant hypotension with the need for inotropes, or requiring surgery (other than vascular site repair), or symptomatic intracranial hemorrhage (ICH), or requiring transfusion of two or three units of red blood cells or equivalent whole blood.
Minor Bleeding:	Bleeding that results in a drop in hemoglobin 3 to <5 g/dL and/or that requires a blood transfusion of 1 unit of red blood cells or equivalent whole blood.
Modified SVS Runoff Score:	Calculated using the angiogram by assessing the patency and degree of stenosis/occlusion in the popliteal artery and the three tibial vessels.[17]
Nonviable Limb:	Condition of extremity (or portion of extremity) in which loss of motor function, neurological function, and tissue integrity cannot be restored with treatment.[2]
Patency:	Patency is defined as a target lesion without a hemodynamically significant stenosis/reocclusion on duplex ultrasound (>50%,) and without target lesion reintervention (TLR).
TransAtlantic Inter-Society Consensus for the Management of Peripheral Arterial Disease (TASC II) Score	Used to classify vessel or arterial disease severity in the aortoiliac and femoropopliteal segments.

16.2 Acronyms

AAA	Abdominal Aortic Aneurysm
ABI	Ankle-Brachial Index
ADE	Adverse Device Effect
AE	Adverse Event
CDT	Catheter-Directed Thrombolysis
CFA	Common Femoral Artery
CFR	Code of Federal Regulations
CIA	Common Iliac Artery
DSMB	Data and Safety Monitoring Board
DUS	Duplex Ultrasound
eCRFs	Electronic Case Report Forms
EC	Ethics Committee
EDC	Electronic Data Capture
EIA	External Iliac Artery
FDA	Food and Drug Administration
GCP	Good Clinical Practice
ICF	Informed Consent Form
IFU	Instructions for Use
IRB	Institution Review Board
ISO	International Standards Organization
ITT	Intent-To-Treat
LAR	Legally Authorized Representative
LE ALI	Lower Extremity Acute Limb Ischemia
LTFU	Lost to Follow-up
PAT	Percutaneous Aspiration Thrombectomy
PMT	Pharmacomechanical Thrombectomy
RHV	Rotating Hemostasis Valve
SAE	Serious Adverse Event
SFA	Superficial Femoral Artery
SOC	Standard of Care
SVS	Society for Vascular Surgery
TASC II	TransAtlantic Inter-Society Consensus for the Management of Peripheral Arterial Disease
TMF	Trial Master File
TIMI	Thrombolysis in Myocardial Infarction
UADE	Unanticipated Adverse Device Event
US	United States
VascuQoL-6	Vascular Quality of Life Questionnaire-6
XTRACT	Aspiration–Based Extraction Technique

17 APPENDIX II: Classifications and Scales

17.1 Rutherford Classification for Acute Limb Ischemia [18, 19]

Category	Description / Prognosis	Findings		Doppler Signal	
		Sensory Loss	Muscle Weakness	Arterial	Venous
I. Viable	Not immediately threatened	None	None	Audible	Audible
IIa. Threatened Marginally	Salvageable if promptly treated	Minimal (toes) or none	None	(Often) Inaudible	Audible
IIb. Threatened Immediately	Salvageable with immediate revascularization	More than toes, associated rest pain	Mild, moderate	(Usually) Inaudible	Audible
III. Irreversible	Major tissue loss or permanent nerve damage inevitable	Profound, anesthetic	Profound, paralysis	Inaudible	Inaudible

17.2 Fontaine Classification [18]

Grade	Symptoms
Stage I	Asymptomatic, incomplete blood vessel obstruction
Stage II	Mild claudication pain in limb
Stage IIA	Claudication at a distance > 200m
Stage IIB	Claudication at a distance < 200m
Stage III	Rest pain, mostly in the feet
Stage IV	Necrosis and/or gangrene of the limb

17.3 Modified SVS Runoff Score [20]

Total Modified SVS Runoff Score is calculated from angiographic images. The score ranges from 1-19, with higher score indicating more severe disease. It is calculated by assessing the patency and degree of stenosis/occlusion in the popliteal artery and the three tibial vessels. The score for the popliteal artery multiplied by 3 and a value of 1 is added before adding all 4 vessel scores together.	
Score (per vessel)	**Level of disease**
0	<20% stenosis
1	21-49% stenosis
2	50-99% stenosis
2.5	Vessel occluded over area < half its length
3	Occlusion > half the vessel length

17.4 TASC II Aortoiliac Lesion Classification [20]

Lesion Type	Description
Type A	Unilateral or bilateral stenosis of Common Iliac Artery (CIA) Unilateral or bilateral single short (≤ 3 cm) stenosis of External Iliac Artery (EIA)
Type B	Short (≤ 3 cm) stenosis of infrarenal aorta Unilateral CIA occlusion Single or multiple stenosis totaling 3-10 cm Involving the EIA not extending into the Common Femoral Artery (CFA) Unilateral EIA occlusion not involving the origins of internal iliac or CFA
Type C	Bilateral CIA occlusions Bilateral EIA stenoses 3-10 cm long not extending into the CFA Unilateral EIA stenosis extending into the CFA Unilateral EIA occlusion that involves the origins of internal iliac and/or CFA Heavily calcified unilateral EIA occlusion, with or without involvement of origins of internal iliac and/or CFA
Type D	Infrarenal aortoiliac occlusion Diffuse disease involving the aorta and both iliac arteries requiring treatment Diffuse multiple stenosis involving the unilateral CIA, EIA, and CFA Unilateral occlusions of both CIA and EIA Bilateral occlusion of EIA Iliac stenosis in patients with Abdominal Aortic Aneurysm (AAA) requiring treatment and not amenable to endograft Placement or other lesions requiring open aortic or iliac surgery

17.5 TASC II Femoral-Popliteal Lesion Classification [20]

Lesion Type	Description
Type A	Single stenosis ≤ 10 cm in length Single occlusion ≤ 5 cm in length
Type B	Multiple lesions (stenoses or occlusion), each ≤5 cm Single stenosis or occlusion ≤ 15 cm not involving the infrageniculate popliteal artery Single or multiple lesions in the absence of continuous tibial vessels to improved inflow for a distal bypass Heavily calcified occlusion ≤ 5 cm in length Single popliteal stenosis
Type C	Multiple stenoses or occlusions totaling > 15 cm, with or without heavy calcifications Recurrent stenoses or occlusions that need treatment after two endovascular interventions
Type D	Chronic total occlusion of the CFA or Superficial Femoral Artery (SFA) (≤20 cm, involving the popliteal artery)

17.6 TIMI Flow [21]

Grade 0	No perfusion
Grade 1	Perfusion past initial occlusion but no distal branch filling.
Grade 2	Perfusion with incomplete or slow distal branch filling.
Grade 3	Full perfusion with filling of all distal branches.

17.7 Vascular Quality of Life – 6 [22]